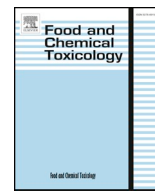




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Short Review

RIFM fragrance ingredient safety assessment, isovaleric acid, CAS Registry Number 503-74-2



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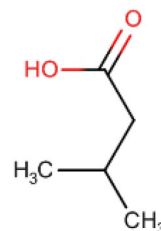
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Version: 091218. This version replaces any previous versions.

Name: Isovaleric acid

CAS Registry Number: 503-74-2



Abbreviation/Definition List:

2-Box Model - A RIFM, Inc. proprietary *in silico* tool used to calculate fragrance air exposure concentration

AF - Assessment Factor

BCF - Bioconcentration Factor

Creme RIFM Model - The Creme RIFM Model uses probabilistic (Monte Carlo) simulations to allow full distributions of data sets, providing a more realistic estimate of aggregate exposure to individuals across a population (Comiskey et al., 2015, 2017; Safford et al., 2015a, 2017) compared to a deterministic aggregate approach

DEREK - Derek Nexus is an *in silico* tool used to identify structural alerts

DST - Dermal Sensitization Threshold

ECHA - European Chemicals Agency

EU - Europe/European Union

GLP - Good Laboratory Practice

IFRA - The International Fragrance Association

LOEL - Lowest Observable Effect Level

MOE - Margin of Exposure

MPPD - Multiple-Path Particle Dosimetry. An *in silico* model for inhaled vapors used to simulate fragrance lung deposition

NA - North America

NESIL - No Expected Sensitization Induction Level

NOAEC - No Observed Adverse Effect Concentration

NOAEL - No Observed Adverse Effect Level

NOEC - No Observed Effect Concentration

NOEL - No Observed Effect Level

OECD - Organisation for Economic Co-operation and Development

OECD TG - Organisation for Economic Co-operation and Development Testing Guidelines

PBT - Persistent, Bioaccumulative, and Toxic

PEC/PNEC - Predicted Environmental Concentration/Predicted No Effect Concentration

QRA - Quantitative Risk Assessment

REACH - Registration, Evaluation, Authorisation, and Restriction of Chemicals

RfD - Reference Dose

RIFM - Research Institute for Fragrance Materials

RQ - Risk Quotient

Statistically Significant - Statistically significant difference in reported results as compared to controls with a $p < 0.05$ using appropriate statistical test

TTC - Threshold of Toxicological Concern

UV/Vis spectra - Ultraviolet/Visible spectra

VCF - Volatile Compounds in Food

VoU - Volume of Use **vPvB** - (very) Persistent, (very) Bioaccumulative

WoE - Weight of Evidence

The Expert Panel for Fragrance Safety* concludes that this material is safe as described in this safety assessment.

This safety assessment is based on the RIFM Criteria Document (Api et al., 2015), which should be referred to for clarifications.

Each endpoint discussed in this safety assessment includes the relevant data that were available at the time of writing (version number in the top box is indicative of the date of approval based on a 2-digit month/day/year), both in the RIFM database (consisting of publicly available and proprietary data) and through publicly available information sources (e.g., SciFinder and PubMed). Studies selected for this safety assessment were based on appropriate test criteria, such as acceptable guidelines, sample size, study duration, route of exposure, relevant animal species, most relevant testing endpoints, etc. A key study for each endpoint was selected based on the most conservative endpoint value (e.g., PNEC, NOAEL, LOEL, and NESIL).

*The Expert Panel for Fragrance Safety is an independent body that selects its own members and establishes its own operating procedures. The Expert Panel is comprised of internationally known scientists that provide RIFM with guidance relevant to human health and environmental protection.

Summary: The existing information supports the use of this material as described in this safety assessment.

Isovaleric acid was evaluated for genotoxicity, repeated dose toxicity, reproductive toxicity, local respiratory toxicity, phototoxicity/photoallergenicity, skin sensitization, and environmental safety. Available data on isovaleric acid and data from read-across analog isobutyric acid (CAS # 79-31-2) show that isovaleric acid is not expected to be genotoxic. Data on isovaleric acid provide a calculated MOE > 100 for the developmental toxicity endpoint. The repeated dose toxicity, fertility, and local respiratory toxicity endpoints were evaluated using the TTC for a Cramer Class I material, and the exposure to isovaleric acid is below the TTC (0.03 mg/kg/day, 0.03 mg/kg/day, and 1.4 mg/day, respectively). The skin sensitization endpoint was completed using available data and the application of the DST for non-reactive materials (900 µg/cm²); exposure is below the DST. The phototoxicity/photoallergenicity endpoints were evaluated based on UV spectra; isovaleric acid is not expected to be phototoxic/photoallergenic. The environmental endpoints were evaluated; isovaleric acid was found not to be PBT as per the IFRA Environmental Standards, and its risk quotients, based on its current volume of use in Europe and North America (i.e., PEC/PNEC), are < 1.

Human Health Safety Assessment

Genotoxicity: Not expected to be genotoxic. (RIFM, 1999; RIFM, 2014a)

Repeated Dose Toxicity: No NOAEL available. Exposure is below the TTC.

Reproductive Toxicity: Developmental Toxicity: NOAEL = 600 mg/kg/day. Fertility: No NOAEL available. Exposure is below the TTC. (ECHA Dossier: Isovaleric acid, 2015)

Skin Sensitization: No safety concerns at current, declared use levels; exposure is below the DST.

Phototoxicity/Photoallergenicity: Not expected to be phototoxic/photoallergenic. (UV Spectra, RIFM Database)

Local Respiratory Toxicity: No NOAEC available. Exposure is below the TTC.

Environmental Safety Assessment**Hazard Assessment:****Persistence:** Screening-level: 3.3 (BIOWIN 3)

(EPI Suite, v4.11; US EPA, 2012a)

Bioaccumulation: Screening-level: 3.16 L/kg

(EPI Suite, v4.11; US EPA, 2012a)

Ecotoxicity: Screening-level: Fish LC50: 382.2 mg/L

(RIFM Framework; Salvito et al., 2002)

Conclusion: Not PBT or vPvB as per IFRA Environmental Standards**Risk Assessment:****Screening-level:** PEC/PNEC (North America and Europe) < 1

(RIFM Framework; Salvito et al., 2002)

Critical Ecotoxicity Endpoint: Fish LC50: 382.2 mg/L

(RIFM Framework; Salvito et al., 2002)

RIFM PNEC is: 0.3822 µg/L

- **Revised PEC/PNECs (2015 IFRA VoU):** North America and Europe: Not applicable; cleared at screening-level

1. Identification

- 1. Chemical Name:** Isovaleric acid
- 2. CAS Registry Number:** 503-74-2
- 3. Synonyms:** Butanoic acid, 3-methyl-; Delphinic acid; 3-Methylbutanoic acid; β-Methylbutyric acid; 3-Methylbutyric acid; 7-メチルノ酸 (C = 4 ~ 30); Isovaleric acid
- 4. Molecular Formula:** C₅H₁₀O₂
- 5. Molecular Weight:** 102.13
- 6. RIFM Number:** 907
- 7. Stereochemistry:** No isomeric center present and no isomers possible.

2. Physical data

- 1. Boiling Point:** 175 °C (FMA Database), 175.25 °C (EPI Suite)
- 2. Flash Point:** 80 °C (GHS), 159 °F; CC (FMA Database)
- 3. Log K_{ow}:** 1.49 (EPI Suite)
- 4. Melting Point:** 3.61 °C (EPI Suite)
- 5. Water Solubility:** 29240 mg/L (EPI Suite)
- 6. Specific Gravity:** 0.937 (FMA Database)
- 7. Vapor Pressure:** 0.798 mm Hg @ 20 °C (EPI Suite v4.0), 0.3 mm Hg @ 20 °C (FMA Database), 1.14 mm Hg @ 25 °C (EPI Suite)
- 8. UV Spectra:** No significant absorbance between 290 and 700 nm; molar absorption coefficient is below the benchmark (1000 L mol⁻¹ · cm⁻¹)
- 9. Appearance/Organoleptic:** [Arctander, Volume II, 1969](#): Colorless liquid; very diffusive, acid-acrid, in moderate dilution cheesy, unpleasant odor of poor tenacity—although the difficulty in removing minute traces from the human skin and the power of the odor give impression of considerable tenacity; in extreme dilution the odor becomes more agreeable, herbaceous, more dry than the odor of n-valeric acid

3. Exposure to fragrance ingredient

- 1. Volume of Use (worldwide band):** 1–10 metric tons per year ([IFRA, 2015](#))
- 2. 95th Percentile Concentration in Hydroalcoholics:** 0.00012% ([RIFM, 2017](#))
- 3. Inhalation Exposure*:** 0.0000054 mg/kg/day or 0.00040 mg/day ([RIFM, 2017](#))
- 4. Total Systemic Exposure**:** 0.000082 mg/kg/day ([RIFM, 2017](#))

*95th percentile calculated exposure derived from concentration survey data in the Creme RIFM Aggregate Exposure Model ([Comiskey et al., 2015](#); [Safford et al., 2015a](#); [Safford et al., 2017](#); and [Comiskey et al., 2017](#)).

**95th percentile calculated exposure; assumes 100% absorption unless modified by dermal absorption data as reported in Section IV. It is derived from concentration survey data in the Creme RIFM Aggregate Exposure Model and includes exposure via dermal, oral, and inhalation

routes whenever the fragrance ingredient is used in products that include these routes of exposure ([Comiskey et al., 2015](#); [Safford et al., 2015a](#); [Safford et al., 2017](#); and [Comiskey et al., 2017](#)).

4. Derivation of systemic absorption

- 1. Dermal:** Assumed 100%
- 2. Oral:** Assumed 100%
- 3. Inhalation:** Assumed 100%

5. Computational toxicology evaluation

- 1. Cramer Classification:** Class I, Low

Expert Judgment	Toxtree v 2.6	OECD QSAR Toolbox v 3.2
I	I	I

- 2. Analogs Selected:**

- a. Genotoxicity:** Isobutyric acid (CAS # 79-31-2)
- b. Repeated Dose Toxicity:** None
- c. Reproductive Toxicity:** None
- d. Skin Sensitization:** None
- e. Phototoxicity/Photoallergenicity:** None
- f. Local Respiratory Toxicity:** None
- g. Environmental Toxicity:** None

- 3. Read-across Justification:** See Appendix below

6. Metabolism

Isovaleric acid is a branched chain aliphatic saturated carboxylic acid, which can be rapidly absorbed from the gastrointestinal tract and metabolized to innocuous products in the body. In an *in vivo* study on Sprague Dawley rats, 4,4-C¹³,1-¹⁴C isovaleric acid administered through feed was completely utilized in the production of cholesterol and fatty acids.

In general, straight or branched chain carboxylic acids undergo β-oxidation. Since isovaleric acid contains a methyl group in position 3, α-oxidation predominates the breakdown into short-chain fragments that can be completely metabolized to carbon-dioxide (CO₂) through acetyl-CoA. It is also established that isovaleric acid is ketogenic and forms a carboxyl moiety and an isopropyl moiety that ultimately undergo cholesterol or fatty acid synthesis.

Isovaleric acid is present as an endogenous metabolite that is converted to acetoacetate and acetyl-CoA and is not expected to accumulate in humans. (JECFA, 1998; HSDB information on Isovaleric acid (accessed on 04/26/2018); ECHA REACH Dossier on Isovaleric acid (accessed on 04/26/2018); [Zabin and Bloch, 1951](#)) (see [Figs. 1 and 2](#)).

Additional References: None.

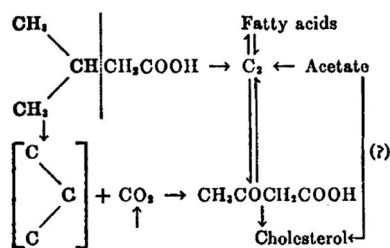


Fig. 1. Metabolism of isovaleric acid in relation to lipid synthesis (Zabin and Bloch, 1951).

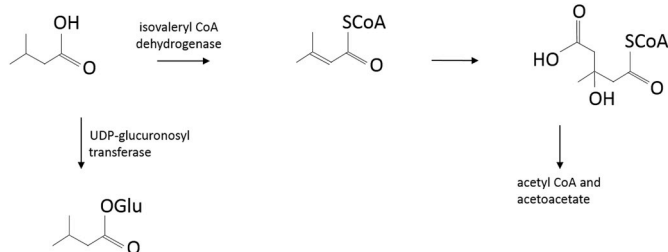


Fig. 2. Metabolism of isovaleric acid via leucine pathway (JECFA, 1998).
Glu- Glucuronic acid; UDP- Uridine 5' - diphosphate.

7. Natural occurrence (discrete chemical) or composition (NCS)

Isovaleric acid is reported to occur in the following foods by the VFC*:

- Anise brandy.
- Apple brandy (calvados).
- Apple fresh (*Malus* species).
- Apple processed (*Malus* species).
- Apricot (*Prunus armeniaca* L.)
- Arctic bramble (*Rubus arcticus* L.)
- Artocarpus species.
- Beef.
- Beer.
- Black currants (*Ribes nigrum* L.)
- Blue cheeses.
- Bread and bread preferment.
- Brown algae.
- Buckwheat.
- Cape gooseberry (*Physalis peruviana* L.)
- Capsicum species.
- Cardamom (*Ellettaria cardamomum* Maton.)
- Cashew apple (*Anacardium occidentale*).
- Cheddar cheese.
- Cheese, various types.
- Cherimoya (*Annona cherimolia* Mill.)
- Cherry (*prunus avium* [sweet], *pr. Cerasus* [sour])
- Chicken.
- Chinese liquor (baijiu).
- Chinese quince (*Pseudocodynia sinensis* Schneid.)
- Cider (apple wine).
- Cinnamomum species.
- Citrus fruits.
- Cocoa category.
- Coffee.
- Crowberry (*Empetrum nigrum* Coll.)
- Elderberry (*Sambucus nigra* L.)
- Filbert, hazelnut (*Corylus avellano*).
- Fish.
- Grape (*Vitis* species).
- Grape brandy.
- Guava and feyoa
- Honey.
- Hop (*humulus lupulus*).
- Katsuobushi (dried bonito).
- Kumazasa (*Sasa albo-marginata*).
- Lamb and mutton.
- Licorice (*Glycyrrhiza* species).
- Litchi (*Litchi chinensis* Sonn.)
- Lobster.
- Loganberry juice (*Rubus ursinus* var. *Loganobaccus*).
- Lovage (*Levisticum officinale* Koch.)
- Maize (*Zea mays* L.)
- Malt.
- Mangifera species.
- Marula (*Sclerocarya birrea* subsp. *Caffra*.)
- Mate (*Ilex paraguayensis*).
- Mentha oils.
- Milk and milk products.
- Mulberry spirit (*Mouro*).
- Mushroom.
- Mussel.
- Mustard (*Brassica* species).
- Oats (*Avena sativa* L.)
- Olive (*Olea europaea*).
- Papaya (*Carica papaya* L.)
- Passion fruit (*Passiflora* species).
- Peach (*Prunus persica* L.)
- Peanut (*Arachis hypogaea* L.)
- Pear brandy.
- Peas (*Pisum sativum* L.)
- Pepper (*Piper nigrum* L.)
- Pineapple (*Ananas comosus*).
- Pistachio nut (*Pistacia vera*).
- Plum brandy.
- Potato (*Solanum tuberosum* L.)
- Potato chips (American).
- Pumpkin seed oil.
- Rapeseed.
- Raspberry, blackberry, and boysenberry.
- Rice (*Oryza sativa* L.)
- Rice cake.
- Rooibos tea (*Aspalathus linearis*).
- Rum.
- Rye bread.
- Sake.
- Salami.
- Sea buckthorn (*Hippophaë rhamnoides* L.)
- Sherry.
- Shoyu (fermented soya hydrolysate).
- Shrimps (prawn).
- Soybean (*Glycine max.* L. Merr.)
- Starfruit (*Averrhoa carambola* L.)
- Strawberry (*Fragaria* species).
- Sugar molasses.
- Sukiyaki.
- Sweet marjoram (*Origanum majorana* L.)
- Swiss cheeses.
- Tarragon (*Artemisia dracunculus* L.)
- Tea.
- Tomato (*Lycopersicon esculentum* Mill.)
- Trassi (cooked).
- Truffle.

Vanilla.
Vinegar.
Wheaten bread.
Whey protein hydrolysate.
Whisky.
Wine.

*VCF Volatile Compounds in Food: database/Nijssen, L.M.; Ingen-Visscher, C.A. van; Donders, J.J.H. (eds). – Version 15.1 – Zeist (The Netherlands): TNO Triskelion, 1963–2014. A continually updated database containing information on published volatile compounds that have been found in natural (processed) food products. Includes FEMA GRAS and EU-Flavis data.

8. IFRA standard

None.

9. REACH dossier

Available; accessed 04/20/18.

10. Summary

10.1. Human health endpoint summaries

10.1.1. Genotoxicity

Based on the current existing data, isovaleric acid does not present a concern for genotoxicity.

10.1.1.1. Risk assessment. Isovaleric acid was assessed in the BlueScreen assay and found positive for both cytotoxicity (positive: < 80% relative cell density) and genotoxicity in the absence of metabolic activation and negative for genotoxicity and cytotoxicity in the presence of metabolic activation (RIFM, 2014b). BlueScreen is a screening assay that assesses genotoxic stress through human-derived gene expression. While the BlueScreen assay on the target material showed positive results, data from additional assays on a more reactive read-across material were considered to fully assess the potential mutagenic or clastogenic effects of the target material.

The mutagenic activity of isovaleric acid has been evaluated in a bacterial reverse mutation assay conducted in compliance with GLP regulations and in accordance with OECD TG 471 using the standard plate incorporation method. *Salmonella typhimurium* strains TA98, TA100, TA1535, TA1537, and TA102 were treated with isovaleric acid in dimethyl sulfoxide (DMSO) at concentrations up to 5000 µg/plate. No increases in the mean number of revertant colonies were observed at any tested concentration in the presence or absence of S9 (RIFM, 1999). Under the conditions of the study, isovaleric acid was not mutagenic in the Ames test.

There are no data assessing the clastogenic activity of isovaleric acid. However, read-across can be made to isobutyric acid (CAS # 79-31-2; see Section V). The clastogenic activity of isobutyric acid was evaluated in an *in vitro* micronucleus test conducted in compliance with GLP regulations and in accordance with OECD TG 487. Human peripheral blood lymphocytes were treated with isobutyric acid in minimal essential medium (MEM) at concentrations up to 880 µg/mL in the presence and absence of metabolic activation (S9) for 4 h and in the absence of metabolic activation for 24 h. Isobutyric acid did not induce binucleated cells with micronuclei when tested up to the maximum concentration in either the presence or absence of an S9 activation system (RIFM, 2014a). Under the conditions of the study, isobutyric acid was considered to be non-clastogenic in the *in vitro* micronucleus test, and this can be extended to isovaleric acid.

Based on the data available, isovaleric acid does not present a concern for genotoxic potential.

Additional References: Heck et al., 1989; RIFM, 1982a; RIFM,

1982b; RIFM, 1983; RIFM, 2015.

Literature Search and Risk Assessment Completed On: 05/09/18.

10.1.2. Repeated dose toxicity

There are insufficient repeated dose toxicity data on isovaleric acid or on any read-across materials. The total systemic exposure to isovaleric acid is below the TTC for the repeated dose toxicity endpoint of a Cramer Class I material at the current level of use.

10.1.2.1. Risk assessment. There are insufficient repeated dose toxicity data on isovaleric acid or on any read-across materials. In a single-dose 90-day subchronic study (testing guidelines unspecified) using male Sprague Dawley rats (6/group), neutralized isovaleric acid (isovaleric acid content: 40% w/w) was administered through the diet at concentrations of 0 (control) or 5% (2500 mg/kg/day). No treatment-related effects were reported in any parameter evaluated. Due to its limitations, this study is used as weight of evidence, and following the JECFA guidelines for the preparation of toxicological working papers on food additives, the NOAEL for 40% isovaleric acid was considered to be 1000 mg/kg/day. (Amoore et al., 1978; data also available at JECFA, 1998; EFSA, 2012; HSDB information on Isovaleric acid; and ECHA Dossier: Isovaleric acid). The total systemic exposure to isovaleric acid (0.082 µg/kg/day) is below the TTC for a Cramer Class I material for the repeated dose toxicity endpoint (30 µg/kg/day; Kroes et al., 2007) at the current level of use.

Additional References: RIFM, 1957 RIFM, 1957.

Literature Search and Risk Assessment Completed On: 07/26/18.

10.1.3. Reproductive Toxicity

The margin of exposure for isovaleric acid is adequate for the developmental toxicity endpoint at the current level of use.

There are insufficient fertility data on isovaleric acid or any read-across materials. The total systemic exposure to isovaleric acid is below the TTC for the fertility endpoint of a Cramer Class I material at the current level of use.

10.1.3.1. Risk assessment. There are sufficient developmental toxicity data on isovaleric acid that can be used to support the developmental toxicity endpoint. In a developmental toxicity study (GLP compliant and similar to OECD 414), which was performed on Wistar rats (10/group), isovaleric acid (99.9% pure) was administered through oral gavage at a dose level of 600 mg/kg/day. All animals were treated during gestation period days 6–19. Salivation in all dams after treatment and local irritation of the larynx and upper and lower respiratory tract were reported; these findings were significant but not severe. No treatment-related effects were reported in any parameter for both dams and fetuses. The NOAEL for maternal, developmental toxicity, and teratogenicity was considered to be 600 mg/kg/day, based on the absence of any treatment-related effects (ECHA Dossier: Isovaleric acid). Since this was a single-dose study with no treatment-related adverse effects observed, a NOAEL of 600 mg/kg/day was considered for the developmental toxicity endpoint. Therefore, the isovaleric acid MOE for the developmental toxicity endpoint can be calculated by dividing the isovaleric acid NOAEL in mg/kg/day by the total systemic exposure to isovaleric acid, 600/0.000082 or 7317073.

In addition, the total systemic exposure to isovaleric acid (0.082 µg/kg/day) is below the TTC (30 µg/kg/day; Kroes et al., 2007; Laufersweiler et al., 2012) for the developmental toxicity endpoint of a Cramer Class I material at the current level of use.

There are insufficient fertility data on isovaleric acid or on any read-across materials that can be used to support the fertility endpoint. The total systemic exposure to isovaleric acid (0.082 µg/kg/day) is below the TTC (30 µg/kg/day; Kroes et al., 2007; Laufersweiler et al., 2012) for the fertility endpoint of a Cramer Class I material at the current level

of use.

Additional References: None.

Literature Search and Risk Assessment Completed On: 08/09/18.

10.1.4. Skin sensitization

Based on existing data and the application of DST, isovaleric acid does not present a safety concern for skin sensitization under the current, declared levels of use.

10.1.4.1. Risk assessment. The chemical structure of this material indicates that it would not be expected to react with skin proteins (Roberts et al., 2007; Toxtree 2.6.13; OECD toolbox v3.4). No predictive skin sensitization studies are available for isovaleric acid. In a human maximization test, no skin sensitization reactions were observed (RIFM, 1977). Acting conservatively, due to the limited data, the reported exposure was benchmarked utilizing the non-reactive DST of 900 µg/cm² (Safford, 2008; Safford et al., 2011; Safford et al., 2015b; Roberts et al., 2015). The current exposure from the 95th percentile concentration is below the DST for non-reactive materials when evaluated in all QRA categories. Table 1 provides the maximum acceptable concentrations for isovaleric acid that present no appreciable risk for skin sensitization based on the non-reactive DST. These concentrations are not limits; they represent acceptable concentrations based on the DST approach.

Additional References: None.

Literature Search and Risk Assessment Completed On: 05/09/18.

10.1.5. Phototoxicity/photoallergenicity

Based on the available UV/Vis spectra, isovaleric acid would not be expected to present a concern for phototoxicity or photoallergenicity.

10.1.5.1. Risk assessment. There are no phototoxicity studies available for isovaleric acid in experimental models. UV/Vis absorption spectra indicate no significant absorption between 290 and 700 nm. The corresponding molar absorption coefficient is well below the benchmark of concern for phototoxicity and photoallergenicity (Henry et al., 2009). Based on lack of absorbance, isovaleric acid does not present a concern for phototoxicity or photoallergenicity.

10.1.5.2. UV spectra analysis. UV/Vis absorption spectra (OECD TG 101) were obtained. The spectra indicate no significant absorbance in

the range of 290–700 nm. The molar absorption coefficient is below the benchmark of concern for phototoxic effects, 1000 L mol⁻¹ · cm⁻¹ (Henry et al., 2009).

Additional References: None.

Literature Search and Risk Assessment Completed On: 04/11/18.

10.1.6. Local Respiratory Toxicity

The margin of exposure could not be calculated due to lack of appropriate data. The exposure level for isovaleric acid is below the Cramer Class I TTC value for inhalation exposure local effects.

10.1.6.1. Risk assessment. There are no inhalation data available on isovaleric acid. Based on the Creme RIFM Model, the inhalation exposure is 0.0004 mg/day. This exposure is 3500 times lower than the Cramer Class I TTC value of 1.4 mg/day (based on human lung weight of 650 g; Carthew et al., 2009); therefore, the exposure at the current level of use is deemed safe.

Additional References: None.

Literature Search and Risk Assessment Completed On: 04/23/18.

10.2. Environmental endpoint summary

10.2.1. Screening-level assessment

A screening-level risk assessment of isovaleric acid was performed following the RIFM Environmental Framework (Salvito et al., 2002), which provides 3 tiered levels of screening for aquatic risk. In Tier 1, only the material's regional VoU, its log K_{ow}, and its molecular weight are needed to estimate a conservative risk quotient (RQ), expressed as the ratio Predicted Environmental Concentration/Predicted No Effect Concentration (PEC/PNEC). A general QSAR with a high uncertainty factor applied is used to predict fish toxicity, as discussed in Salvito et al. (2002). In Tier 2, the RQ is refined by applying a lower uncertainty factor to the PNEC using the ECOSAR model (US EPA, 2012b), which provides chemical class-specific ecotoxicity estimates. Finally, if necessary, Tier 3 is conducted using measured biodegradation and ecotoxicity data to refine the RQ, thus allowing for lower PNEC uncertainty factors. The data for calculating the PEC and PNEC for this safety assessment are provided in the table below. For the PEC, the range from the most recent IFRA Volume of Use Survey is reviewed. The PEC is then calculated using the actual regional tonnage, not the extremes of the range. Following the RIFM Environmental Framework,

Table 1

Maximum Acceptable concentrations for isovaleric acid that present no appreciable risk for skin sensitization based on non-reactive DST.

IFRA Category ^a	Description of Product Type	Maximum Acceptable Concentrations in Finished Products Based on Non-reactive DST	Reported 95th Percentile Use Concentrations in Finished Products
1	Products applied to the lips	0.07%	0.00% ^b
2	Products applied to the axillae	0.02%	0.00% ^b
3	Products applied to the face using fingertips	0.41%	0.00% ^b
4	Fine fragrance products	0.39%	0.00% ^b
5	Products applied to the face and body using the hands (palms), primarily leave-on	0.10%	0.00% ^b
6	Products with oral and lip exposure	0.23%	0.00% ^b
7	Products applied to the hair with some hand contact	0.79%	0.00% ^b
8	Products with significant ano-genital exposure	0.04%	No Data ^c
9	Products with body and hand exposure, primarily rinse-off	0.75%	0.00% ^b
10	Household care products with mostly hand contact	2.70%	0.01%
11	Products with intended skin contact but minimal transfer of fragrance to skin from inert substrate	1.50%	No Data ^c
12	Products not intended for direct skin contact, minimal or insignificant transfer to skin	Not Restricted	0.06%

Note.

^a For a description of the categories, refer to the IFRA/RIFM Information Booklet.

^b Negligible exposure (< 0.01%).

^c Fragrance exposure from these products is very low. These products are not currently in the Creme RIFM Aggregate Exposure Model.

isovaleric acid was identified as a fragrance material with no potential to present a possible risk to the aquatic environment (i.e., its screening-level PEC/PNEC < 1).

A screening-level hazard assessment using EPI Suite v4.11 (US EPA, 2012a) did not identify isovaleric acid as possibly persistent or bioaccumulative based on its structure and physical-chemical properties. This screening-level hazard assessment considers the potential for a material to be persistent *and* bioaccumulative *and* toxic, or very persistent *and* very bioaccumulative as defined in the Criteria Document (Api et al., 2015). As noted in the Criteria Document, the screening criteria applied are the same as those used in the EU for REACH (ECHA, 2012). For persistence, if the EPI Suite model BIOWIN 3 predicts a value < 2.2 and either BIOWIN 2 or BIOWIN 6 predicts a value < 0.5, then the material is considered potentially persistent. A material would be considered potentially bioaccumulative if the EPI Suite model BCFBAF predicts a fish BCF \geq 2000 L/kg. Ecotoxicity is determined in the above screening-level risk assessment. If, based on these model outputs (Step 1), additional assessment is required, a WoE-based review is then performed (Step 2). This review considers available data on the material's physical-chemical properties, environmental fate (e.g., OECD Guideline biodegradation studies or die-away studies), fish bioaccumulation, and higher-tier model outputs (e.g., US EPA's BIOWIN and BCFBAF found in EPI Suite v4.11).

10.2.2. Risk assessment

Based on the current Volume of Use (2015), isovaleric acid presents no risk to the aquatic compartment in the screening-level assessment.

Biodegradation: No data available.

Ecotoxicity: No data available.

Other available data: Isovaleric acid has been registered for REACH with no additional data at this time.

10.2.3. Risk assessment refinement

Ecotoxicological data and PNEC derivation (all endpoints reported in mg/L; PNECs in μ g/L).

Endpoints used to calculate PNEC are underlined.

	LC50 (Fish) (mg/L)	EC50 (<i>Daphnia</i>) (mg/L)	EC50 (Algae) (mg/L)	AF	PNEC (μ g/L)	Chemical Class
RIFM Framework Screening-level (Tier 1)	<u>382.2</u>			1,000,000	0.382	

Exposure information and PEC calculation (following RIFM Environmental Framework: Salvito et al., 2002).

Exposure	Europe (EU)	North America (NA)
Log K_{ow} Used	1.49	1.49
Biodegradation Factor Used	0	0
Dilution Factor	3	3
Regional Volume of Use Tonnage Band	< 1	< 1
Risk Characterization: PEC/PNEC	< 1	< 1

Based on available data, the RQ for this material is < 1. No further assessment is necessary.

The RIFM PNEC is 0.3822 μ g/L. The revised PEC/PNECs for EU and NA are: not applicable. The material was cleared at screening-level and therefore does not present a risk to the aquatic environment at the current reported volumes of use.

Literature Search and Risk Assessment Completed On: 05/02/18.

11. Literature Search

- **RIFM Database:** Target, Fragrance Structure Activity Group materials, other references, JECFA, CIR, SIDS
- **ECHA:** <http://echa.europa.eu/>
- **NTP:** <https://ntp.niehs.nih.gov/>
- **OECD Toolbox**
- **SciFinder:** <https://scifinder.cas.org/scifinder/view/scifinder/scifinderExplore.jsf>
- **PubMed:** <http://www.ncbi.nlm.nih.gov/pubmed>
- **TOXNET:** <http://toxnet.nlm.nih.gov/>
- **IARC:** <http://monographs.iarc.fr>
- **OECD SIDS:** <http://webnet.oecd.org/hpv/ui/Default.aspx>
- **EPA ACToR:** <https://actor.epa.gov/actor/home.xhtml>
- **US EPA HPVIS:** https://ofmpub.epa.gov/opthpv/public_search_publicdetails?submission_id=24959241&ShowComments=Yes&sqlstr=null&recordcount=0&User_title=DetailQuery%20Results&EndPointRpt=Y#submission
- **Japanese NITE:** <http://www.safe.nite.go.jp/english/db.html>
- **Japan Existing Chemical Data Base (JECDB):** http://dra4.nihs.go.jp/mhlw_data/jsp/SearchPageENG.jsp
- **Google:** <https://www.google.com>
- **ChemIDplus:** <https://chem.nlm.nih.gov/chemidplus/>

Search keywords: CAS number and/or material names.

*Information sources outside of RIFM's database are noted as appropriate in the safety assessment. This is not an exhaustive list. The links listed above were active as of 08/27/2018.

Conflicts of interest

The authors declare that they have no conflicts of interest.

Declaration of interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper. We wish to confirm that there are no known conflicts of interest associated with this publication and there has been no significant financial support for this work that could have influenced its outcome. RIFM staff are employees of the Research Institute for Fragrance Materials, Inc. (RIFM). The Expert Panel receives a small honorarium for time spent reviewing the subject work.

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.fct.2019.110570>.

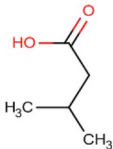
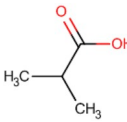
Appendix

Read-across Justification

Methods

The read-across analogs were identified following the strategy for structuring and reporting a read-across prediction of toxicity as described in Schultz et al. (2015). The strategy is also consistent with the guidance provided by OECD within Integrated Approaches for Testing and Assessment (OECD, 2015) and the European Chemical Agency read-across assessment framework (ECHA, 2016).

- First, materials were clustered based on their structural similarity. Second, data availability and data quality on the selected cluster were examined. Third, appropriate read-across analogs from the cluster were confirmed by expert judgment.
- Tanimoto structure similarity scores were calculated using FCFC4 fingerprints (Rogers and Hahn, 2010).
- The physical–chemical properties of the target substance and the read-across analogs were calculated using EPI Suite v4.11 (US EPA, 2012a).
- J_{\max} values were calculated using RIFM's Skin Absorption Model (SAM). The parameters were calculated using the consensus model (Shen et al., 2014).
- DNA binding, mutagenicity, genotoxicity alerts, and oncologic classification predictions were generated using OECD QSAR Toolbox v4.2 (OECD, 2018).
- ER binding and repeat dose categorization were generated using OECD QSAR Toolbox v4.2 (OECD, 2018).
- Developmental toxicity was predicted using CAESAR v2.1.7 (Cassano et al., 2010), and skin sensitization was predicted using Toxtree 2.6.13.
- Protein binding was predicted using OECD QSAR Toolbox v4.2 (OECD, 2018).
- The major metabolites for the target and read-across analogs were determined and evaluated using OECD QSAR Toolbox v4.2 (OECD, 2018).

	Target Material	Read-across
Principal Name	Isovaleric acid	Isobutyric acid
CAS No.	503-74-2	79-31-2
Structure		
Similarity (Tanimoto Score)		0.59
Read-across Endpoint		• Genotoxicity
Formula	C ₅ H ₁₀ O ₂	C ₄ H ₈ O ₂
Molecular Weight	102.13	88.11
Melting Point (°C, EPI Suite)	3.61	-8.29
Boiling Point (°C, EPI Suite)	175.25	153.79
Vapor Pressure (Pa @ 25 °C, EPI Suite)	152	436
Log Kow (KOWWIN v1.68 in EPI Suite)	1.16	0.94
Water Solubility (mg/L, @ 25 °C, WSKOW v1.42 in EPI Suite)	4.07E+004	1.67E+005
Jmax (µg/cm ² /h, SAM)	785.313	3228.890
Henry's Law (Pa·m ³ /mol, Bond Method, EPI Suite)	1.30E-001	9.78E-002
Genotoxicity		
DNA Binding (OASIS v1.4, QSAR Toolbox v3.4)	• No alert found	• No alert found
DNA Binding (OECD QSAR Toolbox v3.4)	• No alert found	• No alert found
Carcinogenicity (ISS)	• Non-carcinogen (moderate reliability)	• Non-carcinogen (moderate reliability)
DNA Binding (Ames, MN, CA, OASIS v1.1)	• No alert found	• No alert found
In Vitro Mutagenicity (Ames, ISS)	• No alert found	• No alert found
In Vivo Mutagenicity (Micronucleus, ISS)	• No alert found	• No alert found
Oncologic Classification	• Not classified	• Not classified
Metabolism		
Rat Liver S9 Metabolism Simulator and Structural Alerts for Metabolites (OECD QSAR Toolbox v4.2)	See Supplemental Data 1	No metabolites

Summary

There are insufficient toxicity data on isovaleric acid (CAS # 503-74-2). Hence, *in silico* evaluation was conducted to determine read-across analogs for this material. Based on structural similarity, reactivity, physical–chemical properties, and expert judgment, isobutyric acid (CAS # 79-31-2) was identified as a read-across material with sufficient data for toxicological evaluation.

Conclusions

- Isobutyric acid (CAS # 79-31-2) was used as a read-across analog for the target material isovaleric acid (CAS # 503-74-2) for the genotoxicity endpoint.

- o The target substance and the read-across analog are structurally similar and belong to the class of branched, saturated organic acids.
- o The only difference between the target substance and the read-across analog is that the read-across analog is 1 carbon smaller compared to the target substance. This structural difference is toxicologically insignificant.
- o Similarity between the target substance and the read-across analog is indicated by the Tanimoto score. Differences between the structures that affect the Tanimoto score are toxicologically insignificant.
- o The physical–chemical properties of the target substance and the read-across analog are sufficiently similar to enable comparison of their toxicological properties.
- o According to the OECD QSAR Toolbox v4.2, structural alerts for toxicological endpoints are consistent between the target substance and the read-across analog.
- o Data described in the genotoxicity section are consistent with *in silico* alerts.
- o The target substance and the read-across analog are expected to be metabolized similarly, as shown by the metabolism simulator.
- o The structural alerts for the endpoints evaluated are consistent between the metabolites of the read-across analog and the target material.

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